Research Article

Elastin Expression is the Strongest Risk Factor for Developing Pelvic Organ Prolapse

Ekspresi Elastin Merupakan Faktor Risiko Terkuat dalam Terjadinya Prolaps Organ Panggul

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Abstract

Objective: To compare elastin expression in the anterior vaginal wall of women with and with no pelvic prolapse.

Methods: The research was conducted in RS Dr. Wahidin Sudirohusodo and other network hospitals of Obstetrics and Gynecology Department of Hasanuddin University, Makassar. Full-thickness specimens were obtained from the anterior vaginal wall of women having a large prolapse repaired (stage III or IV; prolapse group, 34) and the same location in patients with no prolapse having abdominal and vaginal hysterectomy (control group, 35). The expression of elastin was measured by immunohistochemistry on tissue sectioned. The examiner was unaware of sample identity and the patients' clinical history. The result then analyzed with p<0.05 considered significant.

Results: The result of this research shows that with exception of history of bearing baby > 4000 gr weight (p=0.572); age, parity, menopausal status, and body mass index were significantly different between the groups (0.001; 0.035; 0.011; 0.002; respectively). Immunohistochemical staining indicated that elastin expression in the prolapse group was lower (p=0.009). Elastin expression appeared to be stable with increasing of age, parity, menopausal status, history of bearing baby 4000 gr. weight and Body mass Index in the prolapse group. But multiple logistic regression revealed that elastin have the highest influence to prolapse among the risk factors mentioned (Exp.B =6.252).

Conclusion: In this case-control study, the elastin expression were significantly lower in the vaginal wall of patients with a large prolapse. Instead of influence by other risk factors, elastin is the strongest risk factor for developing prolapse among other risk factors. This result is expected to be able to give explanation for the development of prolapse in women without risk factors such is young women and nullipara.

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Keywords: elastin, prolapse, women

Abstrak

***Tujuan**: Membandingkan ekspresi elastin dinding vagina anterior pada perempuan dengan dan tanpa Prolaps organ panggul.*

***Metode**: Penelitian ini dilakukan di RS. Wahidin Sudirohusodo dan jejaringnya Bagian Obstetri dan Ginekologi Universitas Hasanuddin, Makassar. Spesimen jaringan dari dinding vagina anterior pada perempuan dengan prolaps tingkat III dan IV yang menjalani operasi repair (n=34) dan lokasi yang sama pada kelompok kontrol (n=35). Ekspresi elastin diukur dengan menggunakan pemeriksaan immunohistokimia pada jaringan yang telah dipotong. Pemeriksa tidak mengetahui indentitas dan diagnosis klinis sampel. Hasilnya kemudian dianalisis dengan nilai p<0,05 dianggap signifikan.*

***Hasil**: Dengan pengecualian pada riwayat melahirkan bayi dengan BB = 4000 gr, faktor risiko usia, paritas, Indeks Massa Tubuh dan menopause menunjukkan perbedaan yang bermakna antara kelompok prolaps dan kontrol (masing-masing 0,001; 0,035; 0,011; 0,002;). Pemeriksaan Immunohistokimia menunjukkan bahwa ekspresi elastin pada kelompok prolaps secara signifikan lebih rendah (p=0,009). Ekspresi elastin tampaknya tidak dipengaruhi oleh usia, paritas, IMT, menopause dan riwayat melahirkan bayi besar. Tetapi pada analisis regresi logistik multiple menunjukkan bahwa ternyata elastin merupakan faktor yang terkuat yang berperan pada kejadian prolaps dibandingkan faktor risiko lain (Exp.B =6.252).*

***Kesimpulan**: Ekspresi elastin secara signifikan lebih rendah pada kelompok prolaps dan merupakan faktor risiko terkuat untuk menyebabkan prolaps dibandingkan faktor risiko lain. Hal ini diharapkan akan mampu menjelaskan kejadian prolaps pada perempuan tanpa faktor risiko seperti usia muda dan nulipara.*

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Kata Kunci : elastin, faktor risiko, prolaps organ panggul

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INTRODUCTION

Pelvic Organ Prolapse (POP) is define as a pelvic organ protrusion from the vaginal canal. POP is a common condition, causing pressure and activity disturbance in almost 30% of women. it is estimated that approximately 50% of women will lose a supporting mechanism in their pelvic area because of pregnancy and birth process, causing POP in the end.[1]

The highest prevalence of POP is found in advanced age women, thus it is said that the incidence of POP will continue to rise in accordance to longer life expectancy.

The patophysiology of POP itself is still yet to be explained. It is known that the etiology and risk factor of POP is multifactorial, including age, high parity, menopause, obesity and chronically high intra abdominal pressure.[2]

Several studies showed that POP could occur without any risk factors, showing that the quality of a women connective tissue held an important role in its susceptibility of suffering prolapse. Studies on the consistency and composition of the endopelvic fascial tissue and the role of tissue remodeling is now the focus of several ongoing studies.[3]

Connective tissue is constructed of extracellular matrix (ECM), including elastin and collagen as the major protein, and also glycoprotein and proteoglican. The role of elastin, as one of the major structure of ECM in connective tissue, in the pathophysiology of prolapse could be seen in a study performed by Zong dkk. Said study found that trophoelastin, a precursor of elastin, and MMP-9, an enzyme which could degrade elastin, is increased in the patients with POP.[4]

Overall, the high prevalence of POP and its accompanying problems indicate the need of early intervention and identification risk factors, including the elastin in ECM which plays a role in determining the strength of connective tissue in the pathophysiology of prolapse.

This study is expected to show that the elastin expression in ECM of women with POP plays a role in the understanding of the etiology and patophysiology if POP.

METHOD

This study was conducted from January 2011 to April 2012 in several teaching hospitals affiliated with the Department of Obstetrics and Gynecology Faculty of Medicine, University of Hasanudin. The tissue examination was performed in the Pathology Anatomy Laboratorium of Faculty of Medicine University of Hasanudin. This was a cross-sectional study, performed in order to know the difference of elastin expression in women with pelvic organ prolapse and normal women.

The subject of this study was women with severe degree of POP (grade III and IV) who would have an operation and women with benign gynecologic condition who were willing to participate. We explained the objective and method of the study to the subjects and obtained their signature in the consent form. The results of history taking, physical examination and other examination were recorded in a questionnaire.

The tissue sample were fixated with 10% formalin buffer solution. An immunohistochemical examination, with indirect immunoenzyme technique with labeled streptovidin complex, was then performed in the laboratorium. The interpretation of elastin expression was obtained from the histopathological examination of the sample.

The data was then analyzed with chi-square, Fisher exact dan multiple regression test. All data was presented in the form of table or narration. We analyzed the data with SPSS for window versi 17.

RESULT

The study was performed for 1 year and 4 months to patients with POP and patients with benign gynecological tumor in Dr. Wahidin Sudirohusodo hospital and other affiliated hospitals. In this study there were 69 subjects who fulfilled the inclusion criteria. Thirty four subjects were patients with POP as the sample group and 35 subjects were patients with benign gynecological tumor as the control group.

Table 1. The Comparison of Subject's Characteristic in the Case and Control Group

Characteristic	Case group		Control group		p
	n	%	n	%	
Age					
< 45	3	8.8	13	37.1	
45 - 50	4	11.8	9	25.7	0.001
> 50	27	79.4	13	37.1	
Parity					
< 3	11	32.4	21	60.0	
3 - 5	8	23.5	11	31.4	0.035
> 5	15	44.1	3	8.6	
BMI					
Normal	10	31.3	22	68.8	
Overweight	16	59.3	11	40.7	0.011
Obese	8	80.3	2	20.0	
Menopause					
Not yet	4	20	16	80	
< 10 years	10	43.5	13	56.5	0.002
10 -20 years	13	76.5	4	23.5	
> 20 years	7	7	2	22.2	
History of infant > 4000 gr					
Yes	1	33.3	2	66.7	0.572
No	33	50	33	50	

The mean age of the case group was 59.5 years old, significantly higher than in the control group, which was 48.97 years old. in the analysis, age was

further categorized with 45 years old as the cutoff for perimenopause age and 50 years old as the cut-off for menopausal age. Meanwhile, based on analysis with risk approach, the cutoff for parity was three times.

In this study, except for the factor of giving birth to infat weighted > 4000 gr, all risk factors, including age, parity, BMI and the length of menopause showed signficant difference between the group with prolapse and the group without prolapse. This is further explained in Table 1.

Weak elastin expression was found in the 58,8% of subjects in the case group and 25.7% in the control group. Meanwhile, strong elastin expression was found in 31.4% of subjects in the control group but only in 14.7% subjects in the case group. The analysis showed the difference to be significant (p = 0.009).

Table 2. Elastin Expression in the Case Group and Control Group

Group	Elastin expression						p	Linear-by linear association
	Weak		Moderate		Strong			
	n	%	n	%	n	%		
Case	20	58.8	9	26.5	5	14.7	0.019	0.009
Control	9	25.7	15	42.9	11	31.4		

Despite the result mentioned earlier, the expression of elastin did not show significant relation with several risk factors such as age, parity, BMI, menopause and the history of giving birth to infant weighted > 4000 gr in the case group (p=0.556; 0.799; 0.192; 0.971; 0.239, respectively); after performing a multiple logistic regression analysis, we found that elastin possessed the greatest risks (6 times), compared to other risk factors such as age (2.6), parity (1.3), BMI (4.8) and history of giving birth to infant weighted > 4000 gr (1.5).

Table 3. Analysis of each risk Factor of Uterine Prolapse

Risk Factor	B	Exp (B)
Age	.974	2.648
Parity	.242	1.274
Menopause	1.163	3.201
BMI	1.586	4.882
History of giving birth to infant weighted > 4000 gram	.449	1.567
Elastin	1.833	6.252
Constanta	-9.079	.000

DISCUSSION

In this study, it is shown that risk factors such as age, parity, menopause and BMI were significantly different in the group with POP and the group without POP, with the exception of history of giving birth to infant weighted >4000 gr.

The role of age in the etiology of POP evidently has became a general agreement. In a large scale observational study of 971 women, it is found that uterine prolapse, cystocele and rectocele were related to age. Similiar result was also found in this study, in which the mean age of the subject in the case group was significantly higher than in the control group. Both the incidence and the prevalence of POP will increase with age.[5] In a cross sectional study of 1004 women aged 18-83 years old diagnoses with POP for the first time the prevalence increase as much as 40% in each decade.[6] Meanwhile, in another study conducted by Women's Health Initiative (WHI), it is found that women aged 60-69 and 70-79 years old had a higher risk of developing POP compared to women aged 50-59 years old.[7]

Parity also showed a statistically significant relationship with the incidence of prolapse, as often found in most large epidemiological research and observational research, in which the main cause of prolapse were decided on vaginal birth. Pregnancy will cause several changes in the vaginal wall, including the increase of distentibility or the reducing rifidity. Pregnancy also cause a high level of stress to the tissue.[8]

Trauma or other pathology are suspected to cause a change in connective tissue's response to mechanical stress, causing a change in the ECM later on 9. In a study by Tegerstedt et al, it was found that the risk of symptomatic POP increased with the number of children delivered. It is also stated that the women who delivered 4 children had 3.3 times more risks than women who only delivered 1 child. This indicated that stretching and rupture in delivery (both spontaneous rupture or episiotomy) were related to the increasing risk of symptomatic POP.[10] Women's Health Initiative (WHI) stated that a single delivery was related to the increasing risk of uterine prolapse, cystocele, and rectocele.[7] As for the next pregnancy up to the 5 the pregnancy, for each delivery, the risk for prolapse will increase as much as 10-20%.[7] The same finding was also stated in a study by Pelvic Organ Support Study and The Progetto Menopausa.[11]

In a study by Oxford Family Planning, vaginal delivery is the strongest risk factor for the incidence of POP in women older than 60 years old.[12]

Other risk factors for POP include high birthweight, macrosomal baby, long second term of delivery and age younger than 25 years old when delivering the first baby.[13] But in this study, the delivery of macrosomal or baby weighted more than 4000 gram did not have a significant relationship with the incidence of POP (p= 0.572). It differs from other studies, which shows a significant relationship or even a positive relationship between the baby birthweight and the incidence of uterine prolapse.[10,14]

This study showed that BMI had a significant relationship with the incidence of POP. This result is similar with several other studies showing the relationship between obesity and POP. In a study by Women's Health Initiative, BMI >30 kg/m^2 caused an increase in the risk of POP of 40-75%. This was thought to be connected with the increasing of intraabdominal pressure due to the increasing fat tissue in the intraabdominal organ.[15] But, in a study by Washington et al, it is found that obesity actually related more to symptoms worsening instead of the incidence of POP.[16]

In several studies, menopause were indentified as the risk factor for prolapse.[13] But, menopause also means a more advanced age. Due to this fact, it is difficult to decide whether it is anatomical change or aging process due to hormonal factor that plays a role in the occurence of POP.

Ewies et al found that the elastin level did not differ in postmenopausal women receiving hormone replacement therapy (HRT) and women who did not receive HRT.[9] This supports the statement that said once the regeneration process is disturbed, it will not restore to its normal function.

The changes of elastin in POP was not clear until Yamamoto found the level of elastin mRNA and its synthesis underwent a down regulation in the fibroblast culture sample from cardinal ligament of POP patients. The lack of functioning elastin regeneration in adult is a problem, and once the ability to regenerate is lost, restoration to its normal function will not happen.[17]

In this study, it is found that the elastin level in POP patients is significantly lower than in patients without POP (p=0.019). This finding is similar with the result from a study by Goepel et al which found that elastin expression in the sacrouterina ligament is weaker in patients with POP. This shows that elastin plays an important role in the pathophysiology of POP and its lack of expression could be a predisposising factor in the occurrence of POP. A study by Karam et al showed that elastin expression in POP patients is significantly lower than in patients without POP.[18] Study by Goepel and Kantelhardt also found that elastin expression is lacking in the medial and adventitia tunica of uterine artery in patients with POP.[17] But Lin et al had a contrasting finding in their study, in which the elastin expression was found higher in POP patients.[19]

This contrasting results are probably due to the method of the study. The studies used immunohistochemistry method, which depended greatly on the concentration and level of antibody, and the specimen itself. Besides, those studies were employing different staining technique.[18]

In the beginning, it is thought that the decreasing elastin is influenced by risk factors in POP patients such as age, parity, BMI, menopause and the history of giving birth to infant weighted >4000 gr. But after statistical analysis, it turned out that these factors did not have significant relationship with elastin expression.

A risk analysis was performed in this study and it is found that elastin is the strongest risk factor (Exp (B)=6.252) for POP, almost twice compared to menopause and even three times if compared to age. This could explain the insignificant relationship for the previous analysis. It could be said that it is not the risk factors that influencing the level of elastin but rather elastin itself who plays a bigger role in the occurrence of POP. Thus, this study could explain the occurrence of POP in women without apparent risk factors such as in women with young age or women without prior history of vaginal delivery.

The strength of this study is that all samples were processed and assessed randomly by one pathologist who did not know the diagnosis and other data from the sample. Besides, this study also analyzes the relation of each factor to the level of elastin. This study also found the fact that elastin is the strongest factor which can cause POP without the presence of other risk factors. This could explain POP in young women even though this fact is contrasting with the previous theory, which stated that the reduced level of elastin is due to the

aging process or trauma, causing elastin to be more susceptible to proteolytic enzyme such as elastase.

CONCLUSION

From this study, we concluded that elastin expression also plays a role in the occurrence of pelvic organ prolapse. Further studies are still required to understand factors influencing elastin remodeling to prevent the occurrence of POP.

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